experiments with [U-15N,2H,13C]-BsPFK. The assignments allowed for the mapping of peaks representing isoleucine residues onto the crystal structure. This analysis has allowed specific regions of the enzyme involved in the binding of allosteric ligands and the propagation of the allosteric effect to be identified. Funding: NIH-GM33216, NIH-CBI, Welch-A1543.

148-Plat
Microsecond-Resolution Recording of T4 Lysozyme Observes a Brownian Ratchet
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Single-molecule techniques can monitor the kinetics of transitions between enzyme open and closed conformations, but such methods usually lack the resolution to directly observe the underlying transition pathway or any intermediate conformational dynamics. We have recently described a single-molecule *electronic* technique that breaks this barrier (*1-3*). Using a 1 MHz-bandwidth carbon nanotube transistor, the transition dynamics of T4 lysozyme have been recorded with microsecond resolution. We directly resolve a smooth, continuous transition with an average duration of 37 microseconds that suggests a concerted mechanism for glycoside hydrolysis. Unexpectedly, the mechanical closing and re-opening of the enzyme have identical distributions of transition durations, and the motions do not depend on whether the enzyme is in its catalytic or non-productive state. These results illustrate the principle of microscopic reversibility applied to a Brownian ratchet, with lysozyme tracing a single pathway for closing and the reverse pathway for enzyme opening, regardless of its instantaneous catalytic productivity.
1. Y. Choi, *et. al.*, ''Single-Molecule Lysozyme Dynamics Monitored by an Electronic Circuit,'' *Science* **335**, 319 (2012).
2. T.J. Olsen, *et. al.*, ''Electronic Measurements of Single-Molecule Processing by DNA polymerase I (Klenow fragment),'' *JACS* **135**, 7855 (2013).
3. P.C. Sims, *et. al.*, ''Electronic Measurements of Single-Molecule Catalysis by cAMP-Dependent Protein Kinase A,'' *JACS* **135**, 7861 (2013).

149-Plat
Human Neuraminidase Enzymes alter the Lateral Mobility and Function of Integrin Receptors
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Glycolipids and glycoproteins are important components of membrane structure. Mechanisms which alter the structure of glycans at the membrane could influence cellular responses. For example, by removing an important binding epitope or else by unmasking a new one, protein-glycan interactions may be disrupted or reinforced. Our group has been working to understand the role of human neuraminidase enzymes (hNEU) in regulating cellular adhesion and migration through integrin receptors. Of the four hNEU isoenzymes, three have activity at the plasma membrane and lysosome; thus, these enzymes could regulate the composition of the plasma membrane by stripping neuraminic acid (Neu5Ac; also known as sialic acid) from membrane glycans. Using recombinant enzymes and selective hNEU inhibitors developed within our group, we can selectively probe increased or decreased activity of individual isoenzymes in vitro; allowing us to test the effect of specific enzymes. Previous studies have suggested that integrin-mediated adhesion may be altered through hNEU activity. We measured the lateral mobility of integrins in cells treated with NEU3 and NEU4 using single-dye tracking (SDT) by total-internal-reflection fluorescence microscopy (TIRF). We find that hNEU can dramatically change the diffusion of integrin receptors, and that the effect is dependent on the cell type and the isoenzyme used. Adhesion and cell migration assays of cells treated with chemical inhibitors of the enzymes reveal that hNEU activity is intimately involved in the regulation of integrin adhesion to their native ligands. We will present lateral mobility and cell migration assays with enzyme and inhibitor conditions. Our results suggest an important role for hNEU as regulators of membrane composition and the activity of adhesion receptors.

Platform: Membrane Dynamics

150-Plat
Nothing to Sneeze at: A Full-Scale Computational Model of the Human Influenza Virion
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Tackling the ongoing challenge of influenza infectivity would benefit greatly from a molecular understanding of why the influenza A virion exhibits seasonal patterns of infectivity and has wide-ranging survival times in different environments. A computational approach to the study of the behaviour of the virion that focuses on the poorly-understood structural and dynamic role of the lipids is presented here. We have combined experimental data from X-ray crystallography, NMR spectroscopy, cryoelectron microscopy, and lipidomics to build a full-scale computational model of the influenza A virion. This represents the first set of microsecond-scale molecular dynamics simulations of an enveloped virion in explicit solvent that we are aware of. We report results for a set of simulations at different temperatures and with varying lipid compositions. The Forssman glycolipid, which is prevalent in the influenza A lipidome, influences several biophysical characteristics of the virion model, including diffusion and clustering of proteins and lipids. The distribution of peplomers on the virion surface is consistent with accessibility to bivalent antibodies. The distances of binding sites for host cell sialic acid-containing receptors have been analyzed in the virion model for a variety of planar host cell membrane attack orientations. The relatively rigid membrane of the influenza A virion model exhibits a number of biophysical properties consistent with experimental measurements, and may serve as a useful platform for in silico assessment of virion behaviour.

151-Plat
Role Of Membrane-Bending Proteins as Membrane Tension Sensors in Cell Migration
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The plasma membrane (PM) tension has emerged as a key regulator for fundamental cellular functions. However, a missing link between PM tension and biochemical reaction precludes our understanding of how PM tension is coupled to cellular events like directed migration. We found that FBP17, an F-BAR domain protein, acts as a membrane tension sensor that organizes cell polarity during cell migration. The mechanism is based on membrane-bending activity of the F-BAR domain that is counteracted by PM tension. Because FBP17 binds and activates WASP/N-WASP to promote actin polymerization, it corresponds to the local activator of actin polymerization in the feedback loop regulated by PM tension for self-organized formation of the leading edge. These findings provide an important mechanistic insight into cell migration underpinning a wide variety of physiological events such as development, immune response, and cancer metastasis.

152-Plat
Experiment and Simulation Reveal the Bending Properties of Nanoscopic Lipid Domains
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There has great interest in studying the mechanisms that support nanoscopic lipid domain size using model mixtures. To date, experimental studies have focused in large part on structural parameters such as thickness mismatch between phases, lacking access to mechanical properties in direct, experimental ways. The Neutron Spin Echo technique can directly measure the bending modulus of such bilayers systems by observing shape fluctuations. Furthermore, this method permits us to utilize hydrogen-deuterium contrast matching to make direct observations of the mechanical properties of nanoscopic domains themselves.
I will present results detailing for the first time the bending modulus of an individual lipid domain. We have also measured both the structural and mechanical properties of each pure phase compositions of the three component lipid mixture which spontaneously forms these domains. Our results demonstrate that the liquid-ordered phase is more rigid than the liquid disordered phase. Interestingly, the value of the bending modulus in vesicles where these phases coexist as nanoscopic domains approaches that of the liquid ordered-phase, whereas our sample contrast matched so that we only probe the disordered phase in the coexisting system have a bending modulus similar to what is seen in the liquid-disordered only system. We analyze our results with the help of all atom MD simulations which accurately reproduce the structural properties of our system and agree with the trend in mechanical properties that we observe experimentally. These results have implications in determination of important properties such as the line tension which may drive domain size.